ISOENZYME VERIFICATION OF AMERICAN-CHINESE HYBRIDS OF *LIQUIDAMBAR* AND *LIRIODENDRON*[1]

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ABSTRACT

Control-pollinated interspecific hybrids between *Liquidambar styraciflua* and *L. formosana* and between *Liriodendron tulipifera* and *L. chinese* were verified by comparison of isoperoxidase banding patterns developed by starch gel electrophoresis of cambial extracts.

The relationships between some of the trees of eastern Asia and eastern United States (Li, 1952) are especially evident in *Liriodendron,* in which the only two species, the American *Liriodendron tulipifera* L. and *Liriodendron chinese* Helmsley, occur in these regions. There are only four species in *Liquidambar*: *Liquidambar styraciflua* L. occurs in eastern United States, with disjunct populations in Mexico and Central America, and *Liquidambar acalycina* and *Liquidambar formosana* Hance are native to China. A fourth species is found in Turkey.

These two genera were among the first to be investigated after the initiation of a project on cytogenetics, breeding, and improvement of landscape trees at the U.S. National Arboretum in 1967. Santamour (1972a) reported successful crossing among three *Liquidambar* species, using leaf morphology to verify interspecific hybridity. Because verification of young hybrids would be more difficult in *Liriodendron,* in which leaf shape of the two species is very similar, Santamour used the biochemical evaluation of leaf flavonoids (Santamour, 1972b) to establish the true hybridity of seedlings from crosses of *L. chinese* made in 1970. This was thought to be the first controlled hybridization between these species.

However, similar research in China was unknown in the West. According to the Nanjing Technological College of Forest Products (1980), they and the Jiangsu Institute of Botany had successfully crossed the two tuliptrees first in 1963, and then several times between 1963 and 1980. They also reported that the hybrids were superior in growth to *L. chinese* at 12 years of age.

In addition, Huang and Chen (1979) verified both natural and artificial interspecific hybrids by analyses of isoenzyme banding patterns from dormant bud extracts. They found that by acrylamide gel electrophoresis of peroxidase isoenzymes they could distinguish the hybrids that contained the isoenzyme bands of both parents as well as two new bands. Inasmuch as our joint research efforts at the U.S. National Arboretum involved considerable work in isoenzyme electrophoresis, we decided to investigate the potential of using this technique to verify the *Liquidambar* and *Liriodendron* hybrids produced at the U.S. National Arboretum and the *Liriodendron* hybrids produced in China.

MATERIALS AND METHODS

All of the parent trees and the progenies derived from controlled crosses at the National Arboretum were available for study. The first material received from China included dormant twigs from one tree each of the parent species and one putative hybrid. A second shipment contained dormant twigs from three putative hybrids.

Although most isoenzyme studies in plants have utilized leaf tissue, Santamour and Demuth (1980) found that cambial tissue was equally as effective for isoperoxidases. Furthermore, studies of cambial isoenzymes can be made at any time of the year and dormant twigs can be shipped halfway around the world with no adverse effects.

The methods used for starch gel electrophoresis and staining of cambial peroxidase isoenzymes were the same as those reported by Santamour (1982) and have been successful in a wide range of woody genera (Santamour & Demuth, 1980, 1981).

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FIGURE 1. Cambial peroxidase banding patterns in (A) *Liriodendron chinese,* (B) *Liriodendron tulipifera* × *Liriodendron chinese,* (C) *Liriodendron tulipifera,* (D) *Liquidambar formosana,* (E) *Liquidambar styraciflua* × *Liquidambar formosana,* and (F) *Liquidambar styraciflua.*

RESULTS AND DISCUSSION

Isoperoxidase banding patterns of cambial tissue of parent trees and hybrids in *Liquidambar* and *Liriodendron* are shown in Figure 1. The interspecific hybrids showed many, but not all, of the bands found in the parent trees, but the hybrids were easily distinguished. The lack of some parental bands in the hybrids might be expected in view of our lack of knowledge concerning the nature and dominance of the genes involved. Houston and Hood (1982) were able to determine the inheritance patterns of only six of the 13 peroxidase isoenzyme bands they found in leaf tissue of tuliptrees, and those were governed by a single pair of alleles.

With cambial isoperoxidases, it is important that the original parents be used in comparison to the hybrids, and not just an individual of the parent species. We found that it was impossible to verify the tuliptree hybrids created in the People's Republic of China by cambial isoenzymes because the parent trees were not available for study. However, as Huang and Chen (1979) noted, the hybrids could be verified by dormant bud analysis. We also analyzed dormant buds of the Chinese material, and although we could distinguish hybrids, the isoenzyme bands were not very clear.

The use of cambial peroxidase isoenzyme banding patterns is an acceptable method for verifying interspecific hybrids in the cases studied here, especially when material of both parents is also available for study. Our work in *Acer* (Santamour, 1982) and *Quercus* (Santamour & Demuth, 1981) has shown that species belonging to the same infrageneric botanical category, which are potentially sexually compatible, frequently have similar patterns of isoenzyme variation. Thus, an individual of one species may or may not possess a different banding pattern than an individual of another species. Two individuals of the same species, however, may have sufficiently different banding patterns that would allow the detection of intraspecific hybrids between them.

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